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PRELIMINARY STUDIES OF THE EFFECT OF IRRADIATION ON SEEDLING PERFORMANCE OF COWPEA *(Vigna unguiculata (L.) Walp)*.

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Abstract

A study was conducted to assess the effect of six doses of gamma-ray irradiation, viz, 0, 10, 15, 20, 25 and 30 krads on the seedlijng performances of Cornetto and OB89/4 varieties of cowpea which are of Italian and Nigerian origin respectively. The effect of irradiation was observed on germination percentage (G%), days to 50% germiantion (DTG50%). leaf area at 2 weeks (LA2wk), percentage seedling with trifoliate leaves (%STL) and seedling height (SH). There were significant variety X dose interaction for G%, LA2wk and %STL. The overall range of G% and %STL was smaller for Cornetto which had a longer period to attain 50% germination. OB89/4 performed better at low doages (0-15 krads) but Cornetto is more resistant to irradiation at higher dosage (20-30 krads). Using a regression model it was established that Cornetto and OB89/4 required differential irradiation treatment in order to achieve the same effect on seedling traits.

INTRODUCTION

Cowpea *(Vigna unguiculata (L.) Walp)* is of major nutritional importance as food for man and enriches the soil through symbiotic nitrogen fixation. With its high protein content of 22-34% (Bliss *et al.*, 1973; Erskine and Khan, 1976), it complements cereals in the diet of millions of people who cannot affort to produce sufficient animal protein (Singh, 1987). A lot of breedings works aimed at creating superior genotypes (Simmonds, 1989) with the ultimate goal of improving yield (Wilson, 1981) have been carried out in Ngieria. Scully *et al.* (1991) reported that yield was improved by direct selection due to genetic variance and indirect selection for plant ideotype traits affecting yield.

Mutation inducement has been found to be useful as a breeding tool for developing improved legume cultivars (IAEA, 1988). This is currently being employed in the developed world to develop improved cultivars either for genetic or cytogenetic studies (Blixt, 1972; Wellensiek, 1959), disease reistance and agronomic studies in *Phaseolus vulgaris* and *Glycine max* (Tulmann et al; 1988); resistant to pest and disease in *Vigna unguiculata* (Pathak, 1988); yield improvement and seed protein quality in *Cajanus cajan*(Rqavi, 1988). Humphrey, (1954) also found utilizable variations in irradiated *Glycine max* in terms of seed size and oil quality. Although Yu and Yeager, (1960) reported 10 different radiation-induced mutations in tomato which were of no commercial importance, yet they constituted useful genetic markers in tomato breeding. In Nigeria, attention is now focused on the use of irradiation to store food crops. Mutation inducement has been carried out through different methods which include the use of gamma rays; thermal neuron (Jones, 1965) or chemical mutagen (IAEA, 1977). Such mutations were targeted at increasing the genetic variability of cultivars and in nature, they are different from the kind obtainable through gene recombination (Micke, 1988).

Apart from Tulmann *et al*, (1988) who worked on the effect of mutant irradiation on the agronomic characters of beans, Saccardo, (1988) also studied the agronomic performance of pepper *(Capsicum annum)* when irradiated at different stages of the plant growth. He observed that the performance of the crop differed depending on the stage of irradiation and reported uniformity in ripening, adaptability to mechanical harvesting and production of good quality fruits. Cheah (1988) found that 30 krad of gamma irradiation changed the yield components and increased variability of *Phaseolus vulgaris*. He reported an increase of the order of 8 - 15% in seed size and number of pods per plant compared to unirradiated control. He also used five different irradiation doses (5, 10, 15, 20 and 25 krads) on *Arachis hypogea* and found that at the M3 generation, two mutants from the 20 krad treatment showed increases of 32 - 42% in seed size and 1 - 18% in kernel yield per plant compared to unirradiated control.

The use of irradiation to induce mutation for crop improvement is on the increase since it creates an opportunity to evolve novel genotypes. Optimum irradiation dose must be used to give the opportunity for surviving mutants to express themselves phenotypically and enable the breeder to select for desirable mutants. Excessive irradiation doses may have detrimental effect on the embryonic component of the crop thus causing failure in seed germination, emergence and final seedling establishment. The objectives of this preliminary study were to assess the effect of irradiation on the seedling performance of two cultivars of cowpea and to determine the optimum irradiation level for cowpea seedling establishment.

Materials and Methods

Two varieties of cowpea were used in this study. The variety OB89/4 was bred in the Department of Plant Science, Obafemi Awolowo University, Ile-Ife, Nigeria; and the other variety Cornetto is of Italian origin. The seeds of the two varieties dried to 12% moisture content, were irradiated with gamma rays from cobalt 60 at the International Atomic Energy Centre Rome, Italy. The following doses were used: 0, 10, 15, 20, 25 and 30 krads. After the treatment, mutagenic seeds (M1 seeds) were sown in March, 1994 in a randomized complete block design with three replicates in the greenhouse of the Department of Plant Genetics of the University of Naples, Italy. Forty M1 seeds of each variety were planted. Data were collected on the germination percentage (G%) during the first 10 days after planting; days to 50% germination was also estimated, leaf area at 2 - 4 weeks after planting using a leaf area meter, seedling height at 2 - 4 weeks after planting were also obtained. Data were subsequently generated on percentage seedlings with trifoliate (%STL) leaves.

Data taken were subjected to analysis of variance (ANOVA). Genotype means averaged across replications were used in all statistical analyses. The LSD was used for comparison of means at 5% probability level. Regression analysis was performed as described by Steel and Torrie (1980) in order to predict the change due to irradiation.

Results and Discussion

Analysis of Variance (ANOVA).

Table 1 shows the mean square from the ANOVA of the effects of irradiation on seedling traits of the M1 generation of the Cornetto and OB89/4 cowpea varieties. The varieties used were significantly different with respect to days to 50% germination (DT50%), leaf area at 2 weeks (LA 2 weeks) but not for germination percentage G% and percentage seedlings with trifoliate leaves (%STL). The irradiation dosages used had highly significant effects on G%, LA 2 weeks and %STL. Variety X irradiation dosage interaction (V X D) also had signifanct effects on G%, LA 2 weeks but its effect was more pronounced on the %STL. The leaf area measurements for 2, 3 and 4 weeks after planting followed the same trend, hence ANOVA for only LA 2 weeks was presented. The significant treatment effect for G% was largely accounted for by dosage and variety X dosage interaction effects.

The observed mean value of the seedling traits are presented in Table 2 and in Figures 1 and 2. The seedlings of OB 89/4 (V2) established better than that of Cornetto (V1) at low dosages of 0 to 15 krad. At higher dosages of 20 - 30 krads, the Cornetto variety established better than OB89/4. The overall range of irradiation affecting G%, DT50% and %STL were smaller for Cornetto than for OB89/4 (Table 2, and Fig. 1.). It took the Cornetto variety a longer period to attain 50% germiantion than OB89/4. Overall G% values were consistently higher than %STL (Table 2). In this study many more seedlings germinated than were capable of producing trifoliate leaves. This was due to the death of some of the germinated seedlings, others being chlorotic while a few had morphological deformity. Only those that produced trifoliate leaves developed into mature pod-producing plants. It was only in unirradiated plants that %STL was higher than G%. This suggested that a few seeds still emerged after germination count had stopped.

Table 1: Analysis of variance of the effect of irradiation on some seedling traits of two varieties of cowpea.

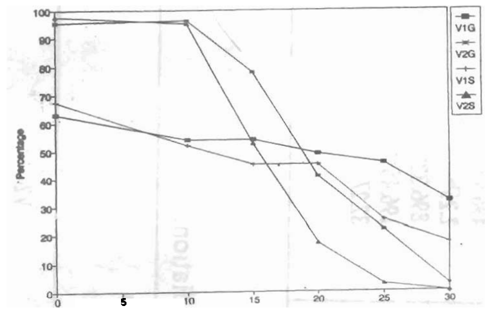
		MEAN SQUARES			
Source of Variation	*DF*	*% Germination (%G)*	*Days to 50% germination (DTG50%)*	*Leaf area at 2 weeks (LA 2wks)*	*Percentage seedlings with trifoliate leaf (%STL)*
Replication	2	31.20^{NS}	3.70NS^{NS}	1.94NS	37.34^{NS}
Treatment	11	378.72**	9.17^{NS}	23.33**	496.92**
Variety (V)	1	44.44^{NS}	75.11**	15.59**	2.25^{NS}
Dosage (D)	5	602.1**	2.51^{NS}	46.37**	896.32**
V X D	5	222.8*	2.65^{NS}	7.34*	196.45**
Error	22	65.95	4.09	2.42	32.27
Total	35				

**, ** = Significant at P = 0.05 and 0.10 levels of probability respectively*
NS = Not Significant

Table 2: Traits of Cowpea seedlings as affected by different doses of gamma irradiation.

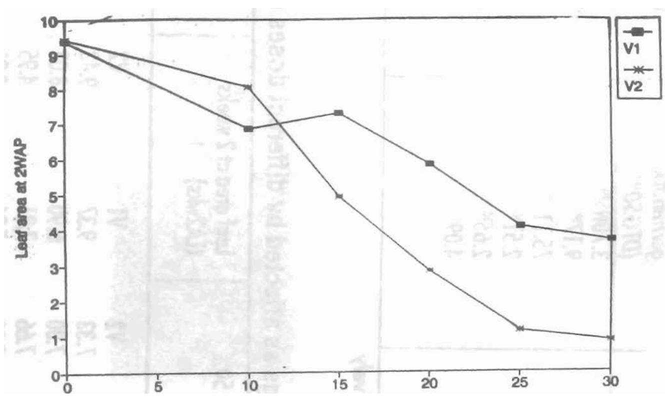
Gamma irradiation (krads)	% Germination Germinatio (%STL)		(%G) Days to 50% (DT50%)		Leaf area at 2 weeks (LA2wks)		Percentage seedlings with trifoliate leaf	
	V1	V2	V1	V2	V1	V2	V1	V2
0	63.03	95.45	9.67	7.33	9.37	9.43	67.5	97.5
10	53.95	96.28	10.33	7.00	6.90	8.08	52.0	95.0
15	53.95	78.02	9.33	7.66	7.31	4.95	45.0	52.5
20	48.97	40.67	9.33	8.00	5.86	2.83	45.0	17.0
25	45.65	21.58	10.33	5.67	4.08	1.16	25.0	2.5
30	31.64	2.49	9.00	5.00	3.70	0.86	17.0	0.0
LSD 0.01%	6.09	5.32	1.52	1.46	1.17	1.08	4.26	3.51

V1 - Cometto, V2 = OB89/4



Levels of irradiations (Krads)

Fig. 1: Effect of irradiation on percentage germination (G) and percentage seedlings with trifoliate leaves (S).



Levels of irradiations (Krads)

Fig 2: Effect of irradiation on leaf area after 2 weeks of planting.

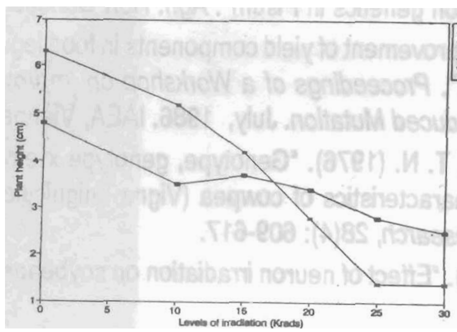
The effect of irradiation on leaf area was more pronounced in OB89/4, than Cornetto (Fig. 2) hence smaller leaf areas were observed in OB89/4. Apparently, Cornetto can withstand the irradiation effects more than OB89/4. Menshal *et al.* (1990) observed that the reduction in the leaf area of mutant cowpeas may be attributed to mutation of the leaf leading to presence or absence of one or two lobes and aberration of leaf form.

The effects of irradiation on the seedling height at 4 weeks is shown in figure 3. The results showed the vulnerability of OB89/4 to high dosage and the relative tolerance of Cornetto to high irradiation doses. Seedlings of OB89/4 were consistently shorter than those of Cornetto especially at the high doses.

Regression analysis

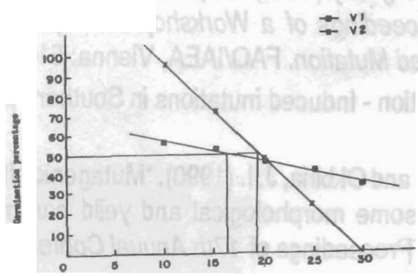
The regressions of germination percentage and %STL on irradiation are shown in Figures 4 and 5 respectively. Regression of G% on irradiation doses showed that OB89/4 is extremely vulnerable to highest dosage used in this study, whereas at that dosage, over 30% germination was still possible in Cornetto. A similar trend was observed for the regression of %STL on irradiation. While no %STL existed for OB89/4 at 30 krads, about

25% of the seedlings of Cornetto still had trifoliate leaves. Figures 4 and 5 showed that different irradiation doses are needed to attain the same result in the two varieties. This confirms the existence of varietal x dosage interaction. For example, at 25 krads (Fig. 5), only 10% and 35% of OB89/4 and Cornetto will have trifoliate leaves. To obtain 50% seedlings with trifoliate leaves, about 16.7 and 18.0 krads of irradiation have to be applied to OB89/4 and Cornetto seeds respectively (Fig. 5). This result also corroborates the earlier observation that Cornetto is more tolerant to irradiation than OB89/4 and also establishes the existence of variety x dosage interaction. Hence differental dosagé treatment are needed in order to bring about comparative effects in these two varieties for mutation.



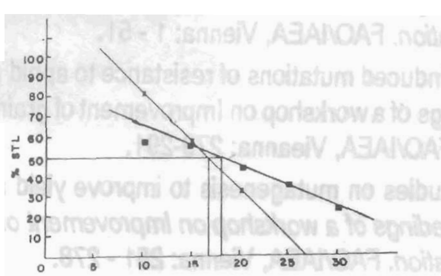
Levels of irradiations (Krads)

Fig. 3: Effect of irradiation on the height of germinated cowpea at 4 weeks after planting.



Dosage (Krads)

Fig. 4: Regression lines of percentage germination on irradiation dosages.



Dosage (Krads)

Fig. 5: Regression lines of percentage seedlings with trifoliate leaf on irradiation dosage.

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